

genetic origin. Claims 43-48 are directed to a kit for transdifferentiating an epidermal basal cell into a cell having one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal or glial cell.

The Office Action, Telephonic Examiner Interview on December 20, 2001, and Applicant's Amendment

In the Final Office Action mailed August 14, 2001, no claims were allowed. Additional acknowledgement was made therein of Applicants' Response to Notice of Non-Compliant Amendment, which Applicants mailed on May 17, 2001, which referred to the remarks filed in Applicants' prior Response to Office Action, which Applicant mailed on March 22, 2001.

On December 20, 2001, Examiner M. Schmidt and Supervisory Patent Examiner J. LeGuyader, graciously granted Applicant's undersigned attorney a telephonic interview with the participation of one of the co-inventors Dr. Toomas Neuman. Applicants again thank the Examiners for their kindness and attention in granting the interview. In the interview, Applicants' undersigned attorney stated Applicants' desire to address the Examiner's grounds of rejection perhaps more clearly than Applicants were able to do in their prior Response to Office Action.

A. Double Patenting Rejections

Claims 1-27 stand rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1-15 of U. S. Patent No. 6,087,168 for the same reasons of record as set forth in the Official Action mailed 11/22/00.

In the Final Office Action, and again in the telephonic interview, on December 20, 2001, the Examiner acknowledged Applicants' stated willingness to file a terminal disclaimer in compliance with 37 C.F.R. 1.321 (c) to overcome an actual or provisional rejection based on a nonstatutory double patenting ground, which Applicants stated in their Response to Office Action, mailed on March 22, 2001.

Applicants submit herewith a terminal disclaimer, since the conflicting application or patent is commonly owned with the above-captioned application.

B. Rejections Under 35 U.S. § 112

Claims 17-39 and 43-48 stand rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described adequately in the specification for the same reasons of record as set forth in the Official Action mailed 11/22/00 as well as the following reasons:

...However, the rejection stands for the scope of such transdifferentiated cells claimed. As argued previously, one skilled in the art would not have been in possession of a representative number of species of such transdifferentiated cells from the teachings of the specification. The specification teaches that may possible outcomes are possible for neuronal type cells exposed to various growth factors and neurotransmitters for instance (see page 5, lines 14-29). It is precisely this variability that creates a broad genus of possible "neuronal-like" cells. The novel feature of the claimed invention is the addition of the antisense oligonucleotides to specific genes in addition to the well-known neural growth factors, which allows the epidermal basal cells to show specific physiological characteristics of a neuronal cell. However, one skilled in the art would not expect such a cell to have all the features of a neuronal cell, nor resemble a neuronal cell which developed naturally in the body since the artificial conditions creates a unique set of gene expression. Thus the scope of possible slight modifications to such a cell is so large that the examples in the specification do not provide a representative number of the possible neuronal-type cells. As such, the claims lack written description as broadly claimed to cells having any possible physiological feature of a neuronal cell...

In response, and as discussed in the telephonic Examiner Interview on December 20, 2001, Applicants have amended Claim 1 to include the subject matter of Claims 12, 14, and 16, which claims are now canceled, without prejudice, merely to avoid redundancy. Thus, amended Claim 1 now includes the recitation of a step (d) which recites a Markush group of signal molecules with which the cell(s) is grown, i.e., "a retinoid compound and a signal molecule selected from the group consisting of BDNF, CNTF, NGF, NT-3, NT-4, IL-6, sonic hedgehog, and sonic hedgehog aminoterminal peptide"; and at least a specified range of markers that are expressed by the transdifferentiated cell(s), i.e., ". . . at least one marker selected from the group consisting of nestin, neural RNA-binding protein Musashi,

neurofilament M, neural-specific β -tubulin, neural-specific enolase, microtubule associated protein 2, glial fibrillary acidic protein (GFAP), O4, or a combination of any of these". The phrase "sonic hedgehog aminoterminal peptide" is recited, while the phrase "functional fragments of any of these" is deleted.

Claim 17, which depends from Claim 1, indirectly includes the additional limitations noted above, making redundant Claim 18, which is now canceled without prejudice. Claim 19 has been amended for greater clarity to recite "... wherein the cell *further* exhibits a lack of mitotic activity under cell culture conditions which induce differentiation in neural progenitor cells ..." so as to conform to amended Claim 1.

Similarly, Claim 43 has been amended to include the recitation of "a retinoid compound and a signal molecule selected from the group consisting of BDNF, CNTF, NGF, NT-3, NT-4, IL-6, sonic hedgehog, and sonic hedgehog aminoterminal peptide." In light of the amendment to Claim 43, Claim 44 has been amended for greater clarity to conform by reference to "(C)", and Claims 46 and 48, now redundant, have been canceled without prejudice. Due to the cancellation of Claim 46, Claim 47 has been amended for clarity to depend directly from Claim 43.

Support for the amendments to Claim 1 and Claim 43 can be found, e.g., in Claims 12, 14, 16, and 48, as originally filed, and at numerous other places in the specification. For instance, support for the recitation of the phrase "signal molecule", instead of the expressions "nerve growth factor" or "neurotrophin", is found in the specification, e.g., at page p.17, lines 2-6. Signal molecules, such as BDNF, CNTF, NGF, NT-3, NT-4, and sonic hedgehog, are disclosed in Claim 12, as originally filed, as well as in the specification, for example, at page 13, line 21 through page 14, line 9; page 16, lines 2-6; page 17, lines 2-6; page 17, lines 12-25; and at page 19, lines 25-28. IL-6 is disclosed in the parent U.S. Patent No. 6,087,168 (originally U.S. Serial No. 09/234,332) at column 6, lines 8-10, the disclosure of which is incorporated by reference in the above-captioned application at page 9, lines 1-2. Support for the recitation of "sonic hedgehog aminoterminal peptide" is found in the specification, e.g., at page 13, line 21 through page 14, line 9. Useful markers are discussed, e.g., at page 14, lines 19-24; page 16, lines 10-19,

and page 17, lines 10-11. The recited expression markers are disclosed in Claim 12, as originally filed, as well as in the specification, e.g., at page 18 lines 8-9 and 18-22.

Moreover, the specification contains extensive and detailed guidance to a wide array of useful markers and other methods for detecting features characteristic of neural progenitors, neuronal and glial cells. For instance, the specifications disclose numerous useful examples of morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell and methods for detecting them such as the discussion of detection methods in the specification at page 14 line 10 through page 17 line 28. While working examples are not required, the specification also discloses working examples of how various neural-specific antigenic markers were detected on the surfaces of transfected cells (specification, page 28 line 10 to page 30 line 4).

Claim 24 has been amended for greater clarity to recite “. . . the morphological feature *comprises* one or more morphological neurite-like process(es) at least about 50 micrometers in length . . .” Claim 13 as originally filed supports this clarifying amendment to open language (“*comprises*”). For greater clarity, Claims 26, 29, 32, 35 and 38 have been amended similary with respect to open language concerning the morphological, physiological and/or immunological features.

In addition, during the telephonic Examiner interview and in response to a question from the the Examiners, Applicants clarified that in accordance with the method of Claim 1, step (b) of Claim 1 renders transfection unnecessary, but that a transfection step could be encompassed within the method of Claim 1.

Additionally, the Examiner requested information regarding BMP antagonists other than fetuin. Support for antagonists of BMP can be found in the specification, for example, at page 4, lines 5-7; page 11, line 24 through page 12, line 11; and page 25, lines 21-24. Applicants' Information Disclosure Statement includes references available at the time the application was filed, pertaining to BMP antagonists, e.g., Reference 26 (Merino *et al.*, Development 126(23):5515-5522 [1999]); Reference 38 (Sela-Donenfeld *et al.*, Development 126(21):4749-62 [1999]); Reference 47 (Zhu *et al.*, Dev. Biol. 215(1):118-29 [1999]); Reference 48 (Zuniga *et al.*, Nature 401(6753):598-602 [1999]); and IDS-cited patent references U.S. Patent No. 5,041,538;

U.S. Patent No. 5,182,375; U.S. Patent No. 5,670,481; U.S. Patent No. 5,679,783; U.S. Patent Nos. 5,821,124; U.S. Patent No. 5,843,775; U.S. Patent No. 5,846,770; and U.S. Patent No. 5,986,056. If the Examiner would like additional references, Applicants would be happy to provide further abstracts regarding known antagonists of BMP.

Applicants believe the amendments herein overcome the ground of rejection. Examiner Schmidt also stated to Applicants' undersigned attorney during the telephonic Examiner Interview that in view of amendments such as Applicants have submitted herewith, she would look toward an allowance. Applicants, therefore, respectfully request the Examiner to withdraw the rejection.

Claims 1-48 stand rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described adequately in the specification for the same reasons of record as set forth in the Official Action mailed 11/22/00 as well as the following reasons:

...In response, the specification as filed teaches on page 14, lines 6-9, that the invention considers administration of the transdifferentiated cells to whole organisms and that once in a whole organism the "plasticity" of the cells would allow the cells to maintain neuronal differentiation "in vitro or in situ, when implanted into the mammalian subject, without the further addition of antisense oligonucleotides." As such, the claims were broadly interpreted to allow for the "detecting the presence or absence of an effect of the potential nerve growth factor" (claim 40 for example) in whole organisms. In other words, wherein the cells are screened *in vivo* after transdifferentiation *in vitro*. Thus the arguments center on the unpredictability of the use of such cells in whole organisms.

The assertion that the specification teaches differentiation of a large subset of the cell population is not questioned, nor is the method for preparing the skin cells for transdifferentiation. The unpredictability in the claims centers on the scope of transdifferentiated cells claimed. As argued above and previously, the scope of the claims reads on making cells having any possible feature of a neuronal cell. Yet the specification as filed teaches specific physiological markers which identify the "transdifferentiated cells" in combination with the methods for making such cells as the novel invention claimed. The specification does not enable one skilled in the art to make and use cells having any possible neuronal feature via the claimed method for the following reasons: (1) the administration of the antisense to specific genes, and the growth of the cells in a specific media form a subset of cells having a specific physiology which differs from other neuronal cells which naturally develop in a whole organism; (2) as the specification teaches, there is a high level of diversity possible in neuronal growth based on the intricacies of numerous factors in the cell in response to the environmental stimuli of the cells. This variability produces a level of

unpredictability in production of any transdifferentiated neuronal cell not enabled by the specification as filed. Hence, the argument that it would take “trial and error” experimentation to make and use the claimed invention primarily centers on the idea that one skilled in the art would not know how to make and use any possible “transdifferentiated cell” with any possible combination of known neuronal growth factors, for instance, as broadly claimed...

As noted above, Applicants have amended Claim 1 to include the subject matter of Claims 12, 14, and 16, which claims are now canceled, without prejudice, merely to avoid redundancy. Thus, amended Claim 1 now includes the recitation of a step (d) which recites a Markush group of signal molecules with which the cell(s) is grown, i.e., “a retinoid compound and a signal molecule selected from the group consisting of BDNF, CNTF, NGF, NT-3, NT-4, IL-6, sonic hedgehog, and sonic hedgehog aminoterminal peptide”; and at least a specified range of markers that are expressed by the transdifferentiated cell(s), i.e., “ . . . at least one marker selected from the group consisting of nestin, neural RNA-binding protein Musashi, neurofilament M, neural-specific β -tubulin, neural-specific enolase, microtubule associated protein 2, glial fibrillary acidic protein (GFAP), O4, or a combination of any of these”.

Also regarding Claims 1-48, the Examiner noted that claim amendments in Applicant’s Response to the prior Office Action clarified the negative regulators claimed but the Examiner asserted that the claims as written “do not specify *in vitro* and thus still broadly read on *in vivo* use.” Applicants have amended the preambles of Claims 1, 29, and 43, to recite “*in vitro*” or “cultured *in vitro*” to clarify the *in vitro* nature of the claimed invention.

Applicant has canceled Claims 40, 41, and 42, without prejudice. Therefore, the ground of rejection is mooted concerning Claims 40, 41, and 42, directed to *in vitro* screening or assay methods, e.g., for potential new drugs. The cancellation of Claims 40, 41, and 42 is made with the reservation of Applicants’ right to pursue the subject matter of these claims in a continuation application, which right was acknowledged by the Examiner during the interview.

Applicants believe that in view of Applicants’ amendments, Claims 1-11, 13, 15, 17, 19, 20, 22-39, 43, 44, 45, and 47 are ready for allowance. Examiner Schmidt stated to Applicants’ undersigned attorney during the telephonic Examiner Interview, on December 20,

2001, that in view of such amendments, which were then proposed, she would look toward an allowance.

CONCLUSION

In view of the above amendments and remarks and as indicated by the Examiner during the interview by the Examiner, it is submitted that this application is now ready for allowance. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned attorney at (213) 896-6665.

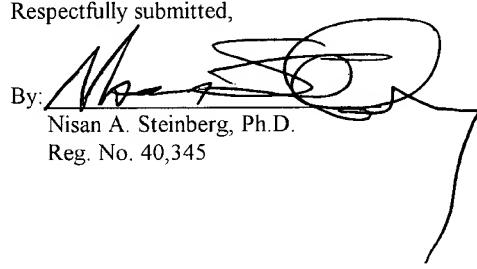
CONCLUSION

In view of the above amendments and remarks, it is submitted that this application is now ready for allowance. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned attorney at (213) 896-6665.

Respectfully submitted,

By:

Nisan A. Steinberg, Ph.D.
Reg. No. 40,345



SIDLEY AUSTIN BROWN & WOOD
555 West Fifth Street
Los Angeles, California 90013
Ofc: 213/ 896-6665
Fax: 213/ 896-6600

Version with Markings to Show Changes Made

In the Claims:

Please cancel Claims 12, 14, 16, 18, 21, 40, 41, 42, 46, and 48 without prejudice.

Please amend Claims 1, 19, 24, 26, 29, 32, 35, 38, 43, and 47 as follows.

1. (Twice Amended) An in vitro method of transdifferentiating an epidermal basal cell into a cell having one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell, comprising:

(a) culturing a proliferating epidermal basal cell population comprising one or more epidermal basal cell(s), said cell(s) derived from the skin of a mammalian subject;

(b) exposing the cell(s) to an amount of an antagonist of bone morphogenetic protein (BMP) effective to antagonize endogenous BMP signal transduction activity; [and]

(c) growing the cell(s) in the presence of at least one antisense oligonucleotide comprising a segment of a human MSX1 gene and/or a segment of a human HES1 gene, or homologous non-human counterpart of either of these, in an amount effective to suppress the expression of functional gene product of MSX1 and/or HES1, whereby the cell is transdifferentiated into a cell having one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell[.]; and

(d) growing the transdifferentiated cell in a medium comprising a retinoid compound and a signal molecule selected from the group consisting of brain-derived neurotrophic factor (BDNF), ciliary neurotrophic factor (CNTF), platelet-derived growth factor (PDGF), nerve growth factor (NGF), neurotrophin (NT)-3, neurotrophin (NT)-4, IL-6, sonic hedgehog, and sonic hedgehog aminoterminal peptide, and wherein the physiological and/or immunological feature comprises expression of a marker selected from the group consisting of nestin, neural RNA-binding protein Musashi, neurofilament M, neural-specific β-tubulin, neural-specific enolase, microtubule associated protein 2, glial fibrillary acidic protein (GFAP), O4, or a combination of any of these.

2. (Reiterated) The method of Claim 1, wherein the subject is a human.
3. (Reiterated) The method of Claim 1, wherein the epidermal basal cell(s) is derived from a skin biopsy.
4. (Reiterated) The method of Claim 1, wherein culturing the proliferating epidermal basal cell population further comprises separating keratinized epidermal cells from the epidermal basal cells in a calcium-free medium.
5. (Reiterated) The method of Claim 1, wherein the amount of the antagonist of bone morphogenetic protein is about 10^{-6} to 10^{-4} M.
6. (Reiterated) The method of Claim 5, wherein the amount of the antagonist of bone morphogenetic protein is about 5×10^{-6} to 5×10^{-5} M.
7. (Reiterated) The method of Claim 1, wherein the antagonist of bone morphogenetic protein (BMP) is fetuin, noggin, chordin, gremlin, or follistatin.
8. (Reiterated) The method of Claim 7, wherein the fetuin is mammalian or avian fetuin.
9. (Reiterated) The method of Claim 8, wherein the mammalian fetuin is human, bovine, porcine, ovine, or equine fetuin.

10. (Reiterated) The method of Claim 1, wherein the antisense oligonucleotide(s) is modified with one or more thio groups.

11. (Reiterated) The method of Claim 1, wherein the amount of the antisense oligonucleotide is about 5×10^{-6} M to about 10^{-5} M.

12. Canceled.

13. (Reiterated) The method of Claim 1, wherein the morphological feature comprises one or more morphological neurite-like process(es) at least about 50 micrometers in length.

14. Canceled.

15. The method of Claim 14, wherein the retinoid compound is all-trans retinoic acid or Vitamin A.

16. Canceled.

17. (Reiterated) A transdifferentiated cell of epidermal origin having one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell produced by the method of Claim 1.

18. Canceled.

19. (Amended) The transdifferentiated cell of Claim 17, wherein the cell further exhibits a lack of mitotic activity under cell culture conditions which induce differentiation in neural progenitor cells.

20. (Reiterated) The cell of Claim 17, wherein the transdifferentiated cell has a morphological, physiological and/or immunological feature specific to a neuronal cell.

21. Canceled.

22. (Reiterated) The transdifferentiated cell of Claim 20, wherein the cell is a GABAergic cell.

23. (Reiterated) The transdifferentiated cell of Claim 20, wherein the cell is a dopaminergic cell.

24. (Amended) The transdifferentiated cell of Claim 17, wherein the morphological feature comprises [is] one or more neurite-like process(es) at least about 50 micrometers in length.

25. (Reiterated) The cell of Claim 17, wherein the transdifferentiated cell has a morphological, physiological, or immunological feature specific to an astroglial or oligodendroglial cell.

26. (Amended) The transdifferentiated cell of Claim 25, wherein the immunological feature comprises [is] expression of glial fibrillary acidic protein (GFAP) or O4.

27. (Reiterated) The transdifferentiated cell of Claim 17, wherein the cell is of human origin.

28. (Reiterated) A cell culture derived from the transdifferentiated cell of Claim 17, comprising a plurality of cells that express one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell.

29. (Amended) A transdifferentiated cell of epidermal origin and cultured in vitro, comprising a cell of epidermal basal cell origin, said transdifferentiated cell displaying one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell, wherein the physiological and/or immunological feature comprises [is] expression of a marker selected from the group consisting of nestin, neural RNA-binding protein Musashi, neurofilament M, neural-specific β -tubulin, neural-specific enolase, microtubule associated protein 2, glial fibrillary acidic protein (GFAP), O4, or a combination of any of these.

30. (Reiterated) The transdifferentiated cell of Claim 29, wherein the cell further displays the physiological feature of a lack of mitotic activity under cell culture conditions which induce differentiation in neural progenitor cells.

31. (Reiterated) The cell of Claim 29, wherein the transdifferentiated cell has a morphological, physiological, or immunological feature specific to a neuronal cell.

32. (Amended) The transdifferentiated cell of Claim 31, wherein the physiological and/or immunological feature comprises [is] expression of neural RNA-binding

protein Musashi, neurofilament M, neural-specific β -tubulin, neural-specific enolase, microtubule associated protein 2.

33. (Reiterated) The transdifferentiated cell of Claim 31, wherein the cell is a GABAergic cell.

34. (Reiterated) The transdifferentiated cell of Claim 31, wherein the cell is a dopaminergic cell.

35. (Amended) The transdifferentiated cell of Claim 29, wherein the morphological feature comprises [is] one or more neurite-like process(es) at least about 50 micrometers in length.

36. (Reiterated) The transdifferentiated cell of Claim 29, wherein the cell is of human origin.

37. (Reiterated) The cell of Claim 29, wherein the transdifferentiated cell has a morphological, physiological, or immunological feature specific to an astroglial or oligodendroglial cell.

38. (Amended) The transdifferentiated cell of Claim 37, wherein the physiological and/or immunological feature comprises [is] expression of glial fibrillary acidic protein (GFAP) or O4.

39. (Reiterated) A cell culture derived from the transdifferentiated cell of Claim 29, comprising a plurality of cells that express one or more morphological, physiological

and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell.

40. Canceled.

41. Canceled.

42. Canceled.

43. (Amended) A kit for transdifferentiating, in vitro, an epidermal basal cell into a cell having one or more morphological, physiological and/or immunological feature(s) of a neural progenitor, neuronal, or glial cell, comprising:

(A) an antagonist of bone morphogenetic protein (BMP); [and]
(B) at least one antisense oligonucleotide comprising a segment of a human MSX1 gene, a segment of a human HES1 gene, or a non-human homologous counterpart of either of these[.]; and

(C) a retinoid compound and a signal molecule selected from the group consisting of brain-derived neurotrophic factor (BDNF), ciliary neurotrophic factor (CNTF), platelet-derived growth factor (PDGF), nerve growth factor (NGF), neurotrophin (NT)-3, neurotrophin (NT)-4, IL-6, sonic hedgehog, and sonic hedgehog aminoterminal peptide.

44. (Reiterated) The kit of Claim 43, further comprising instructions for using (A),[and] (B), and (C) in transdifferentiating a subject's epidermal basal cell(s).

45. (Reiterated) The kit of Claim 43, wherein the antagonist of bone morphogenetic protein (BMP) is fetuin, noggin, chordin, gremlin, or follistatin.

46. Canceled.

47. (Amended) The kit of Claim [46]43, wherein the retinoid compound is all-trans retinoic acid or Vitamin A.

48. Canceled.